WE CLAIM:

- 1. A sensor for binding a molecule in an aqueous sample in an assay, the sensor comprising:
 - a substrate surface;
- a layer on the substrate surface comprising polymer chains having two termini and a water-soluble or water-dispersible intermediate segment between the termini, one terminus being free and the other terminus being bound to the substrate surface, the intermediate segment containing groups for the attachment of a probe for binding the molecule, said groups being capable of attaching said probe without first being subjected to a chemical treatment to activate said for probe attachment; and,
 - a probe for binding the molecule.
- 2. The sensor of claim 1 wherein a hydrophilic layer comprises the water-soluble or water-dispersible polymer chain segments, said segments comprising repeat units derived from a water-soluble or water-dispersible monomer having a log P value of less than about 1.
- 3. The sensor of claim 1 wherein a hydrophilic layer comprises the water-soluble or water-dispersible polymer chain segments, said segments comprising repeat units derived from a water-soluble or water-dispersible monomer having a log P value of less than about 0.1.
- 4. The sensor of claim 1 wherein said water-soluble or water-dispersible intermediate segment is formed from a monomer having the formula:

wherein P comprises a functional group capable of undergoing free radical polymerization, E comprises a functional group capable of reacting with the probe, m is greater than or equal to 0, and X, when present, is a moiety which links P and X.

5. The sensor of claim 4 wherein said water-soluble or water-dispersible intermediate segment is formed from a monomer having the formula:

wherein: R⁴ is a hydrogen or an alkyl substitutent; each R⁷ is selected from the group consisting of hydrogen, substituted or unsubstituted hydrocarbyl, heterohydrocarbyl, alkoxy, substituted or unsubstituted aryl, sulphates, and thioethers; X, when present, is a linking moiety selected from the group consisting of substituted or unsubstituted hydrocarbylene and heterohydrocarbylene; m is greater than or equal to 0; and, w is an integer ranging from about 1 to 4.

6. The sensor of claim 5 wherein m is 0, the monomer having the formula:

128

$$\begin{array}{c}
R^4 \\
I \\
I \\
C = C \\
C = O \\
O \\
O \\
(R^7)_{W}
\end{array}$$

wherein R⁴, R⁷ and w are as defined in claim 4.

7. The sensor of claim 5 wherein m is an integer ranging from about 1 to 5, X being a linking moeity represented by the formula:

$$\begin{array}{c|c} C & \\ \hline - C & \\ \hline - N & \\ \hline - C & \\ H & \end{array} \begin{array}{c} C \\ \hline - N \\ \hline - C & \\ \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \hline - N \\ \hline \end{array} \begin{array}{c} C \\ \end{array} \end{array} \begin{array}{c} C \\ \end{array} \begin{array}{c} C \\ \end{array} \begin{array}{c} C \\ \end{array} \begin{array}{c} C \\ \end{array} \end{array} \begin{array}{c} C \\ \end{array} \begin{array}{c} C \\ \end{array} \begin{array}{c} C \\ \end{array} \begin{array}{c} C \\ \end{array} \end{array} \begin{array}{c} C \\ \end{array} \end{array} \begin{array}{c} C \\ \end{array} \begin{array}{c} C \\ \end{array} \begin{array}{c} C \\ \end{array} \begin{array}{c} C \\ \end{array} \end{array} \begin{array}{c} C \\ \end{array} \begin{array}{c} C \\ \end{array} \end{array} \begin{array}{c$$

wherein n is an integer ranging from about 1 to 5.

8. The sensor of claim 7 wherein said water-soluble or water-dispersible intermediate segment is formed from a monomer selected from group consisting of:

$$\begin{array}{c}
R^{4} \\
I \\
C = C \\
C = O \\
NH \\
(CH_{2})_{n} \\
C = O \\
O \\
N
\end{array}$$

$$\begin{array}{c}
C = O \\
O \\
O \\
O \\
(R^{7})_{w}
\end{array}$$

and

wherein R^4 , R^7 and n are as defined in claim 7.

9. The sensor of claim 4 wherein said water-soluble or water-dispersible intermediate segment is formed from a monomer selected from the group consisting of:

$$= \bigvee_{\mathbb{R}^9} \bigvee_{\mathbb{R}^9} \bigvee_{\mathbb{R}_9} \bigvee_{\mathbb{R}$$

wherein R⁹ is hydrogen or hydrocarbyl.

- 10. The sensor of claim 9 wherein said water-soluble or water-dispersible intermediate segment is formed from a monomer selected from the group consisting of N-acryloxysuccinimide, 2-(methylacryloyloxy)ethyl acetoacetate, glycidyl methacrylate and 4,4-dimethyl-2-vinyl-2-oxazolin-5-one.
- 11. The sensor of claim 1 wherein the polymer chains additionally comprise a hydrophobic segment between the water-soluble or water-dispersible intermediate segment and the substrate.
- 12. The sensor of claim 11 wherein the hydrophobic segment has a dry thickness ranging from about 50 to about 2000 angstroms.
- 13. The sensor of claim 11 wherein the hydrophobic segment has a dry thickness ranging from about 100 to about 1500 angstroms.

- 14. The sensor of claim 11 wherein, in addition to said hydrophobic polymer chain segments, spacer molecules are also attached to the substrate surface, the ratio of said hydrophobic polymer chain segments to the sum of hydrophobic polymer chains segments and spacer molecules ranging from about 0.2 to about 0.8.
- 15. The sensor of claim 11 wherein the hydrophobic polymer chain segments are linear.
- 16. The sensor of claim 11 wherein the hydrophobic polymer chain segments are branched.
- 17. The sensor of claim 11 wherein at least a portion of the hydrophobic polymer chain segments are crosslinked to other hydrophobic polymer chain segments in the hydrophobic layer.
- 18. The sensor of claim 11 wherein the ratio of water-soluble or water-dispersible segments to hydrophobic segments is less than about 0.8:1.
- 19. The sensor of claim 11 wherein the hydrophobic layer comprises hydrophobic polymer chain segments, one end of each of said hydrophobic segments being attached to the substrate surface, and further wherein a hydrophilic layer comprising the water-soluble or water-dispersible polymer chain segments, one end of each of said water-soluble or water-dispersible segments being attached to the hydrophobic layer, the ratio of water-soluble or water-dispersible segments to a number of sites of attachment on the substrate surface being less than about 1:1.
- 20. The sensor of claim 20 wherein the ratio of water-soluble or water-dispersible segments to the number of sites of attachment on the substrate surface is less than about 0.8:1.

- 21. The sensor of claim 11 wherein said hydrophobic polymer chain segment comprises repeat units derived from a hydrophobic monomer having a log P value of at least about 0.5.
- 22. The sensor of claim 11 wherein said hydrophobic polymer chain segment comprises repeat units derived from a hydrophobic monomer having a log P value of at least about 1.
- 23. The sensor of claim 11 wherein in an absolute difference between a log P value of the hydrophobic monomers and the water-soluble or water-dispersible monomers, from which said water-soluble or water-dispersible segment is derived, is at least about 1.
- 24. The sensor of claim 1 wherein the water-soluble or water-dispersible polymer chain segments have sites for the attachment of a probe, and a weight average molecular weight of at least 1,000 but no more than 5,000,000.
- 25. The sensor of claim 1 wherein a hydrophilic layer comprising the water-soluble or water-dispersible segments has a dry thickness ranging from about 10 angstroms to about 2000 angstroms.
- 26. The sensor of claim 1 wherein the molecule is a biological molecule and the probe is selected from the group consisting of nucleic acids, polypeptides, peptide nucleic acids, markers, cells, elastin, collagen, carbohydrates, enzymes, lipids, phospholipids, hormones, drug targets, phosphates, and metal ions.
- 27. The sensor of claim 1 wherein said functionalized groups are selected from the group consisting of hydroxy groups, amino groups, carboxylic acids, carboxylic acid derivatives, and thiols.

- 28. The sensor of claim 1 wherein the intermediate segment is substantially free of crosslinks to the intermediate segment of other polymer chains.
- 29. A sensor for binding a molecule in an aqueous sample in an assay, the sensor comprising:
 - a substrate surface;

a layer on the substrate surface comprising polymer chains having two termini and a water-soluble or water-dispersible intermediate segment between the termini, one terminus being free and the other terminus being bound to the substrate surface, the intermediate segment comprising a residue of a monomer having a probe for binding the molecule attached thereto.

- 30. The sensor of claim 29 wherein the probe and monomer have a size ratio which is less than about 50:1 (probe:monomer).
- 31. The sensor of claim 29 wherein the probe and monomer have a size ratio which is less than about 10:1 (probe:monomer).
- 32. The sensor of claim 29 wherein the probe and monomer have a size ratio which is less than about 5:1 (probe:monomer).
- 33. The sensor of claim 29 wherein the molecule is a biological molecule and the probe is selected from the group consisting of nucleic acids, polypeptides, peptide nucleic acids, markers, cells, elastin, collagen, carbohydrates, enzymes, lipids, phospholipids, hormones, drug targets, phosphates, and metal ions.
- 34. The sensor of claim 33 wherein the probe is a nucleic acid polymer.

- 35. The sensor of claim 34 wherein the nucleic acid probe is a DNA, RNA or PNA fragment, or a derivative thereof.
- 36. The sensor of claim 34 wherein the probe is at least about 5 nucleotides in length.
- 37. The sensor of claim 36 wherein the probe is about 10 to 100 nucleotides in length.
- 38. The sensor of claim 29 wherein the polymer chains additionally comprise a hydrophobic segment between the water-soluble or water-dispersible intermediate segment and the substrate.
- 39. The sensor of claim 38 wherein the hydrophobic segment has a dry thickness of at least about 50 to about 2000 angstroms.
- 40. The sensor of claim 38 wherein, in addition to said hydrophobic polymer chain segments, spacer molecules are also attached to the substrate surface, the ratio of said hydrophobic polymer chain segments to the sum of hydrophobic polymer chains segments and spacer molecules ranging from about 0.2 to about 0.8.
- 41. The sensor of claim 38 wherein at least a portion of the hydrophobic polymer chain segments are crosslinked to other hydrophobic polymer chain segments in the hydrophobic layer.
- 42. The sensor of claim 38 wherein the ratio of water-soluble or water-dispersible segments to hydrophobic segments is less than about 0.8:1.
- 43. The sensor of claim 38 wherein the hydrophobic layer comprises hydrophobic polymer chain segments, one end of each of said hydrophobic segments being attached to the substrate surface, and further wherein a hydrophilic layer comprising the water-soluble or water-dispersible polymer

chain segments, one end of each of said water-soluble or water-dispersible segments being attached to the hydrophobic layer, the ratio of water-soluble or water-dispersible segments to a number of sites of attachment on the substrate surface being less than about 1:1.

- 44. The sensor of claim 38 wherein said hydrophobic polymer chain segment comprises repeat units derived from a hydrophobic monomer having a log P value of at least about 0.5.
- 45. The sensor of claim 44 wherein in the absolute difference between the log P value of the hydrophobic monomers and the water-soluble or water-dispersible monomers, from which said water-soluble or water-dispersible segment is derived, is at least about 1.
- 46. The sensor of claim 29 wherein the water-soluble or water-dispersible polymer chain segments have groups for the attachment of a probe, and a weight average molecular weight of at least 1,000 but no more than 5,000,000.
- 47. The sensor of claim 29 wherein a hydrophilic layer comprising the water-soluble or water-dispersible segments has a dry thickness ranging from about 10 angstroms to about 2000 angstroms.
- 48. The sensor of claim 29 wherein the intermediate segment is substantially free of crosslinks to the intermediate segment of other polymer chains.
- 49. A polymer brush for binding a molecule in an aqueous sample in an assay, the brush comprising a substrate surface, a hydrophobic layer comprising hydrophobic polymer chain segments attached to the substrate surface and having a dry thickness of at least about 50 angstroms, and a hydrophilic layer attached to the hydrophobic layer containing functional groups for the attachment of a probe for binding the molecule.

- 50. The polymer brush of claim 49 wherein the hydrophobic layer has a dry thickness of at least about 100 angstroms.
- 51. The polymer brush of claim 49 wherein the hydrophobic layer has a dry thickness of at least about 1000 angstroms.
- 52. The polymer brush of claim 49 wherein the hydrophobic layer has a dry thickness of at least about 2000 angstroms.
- 53. The polymer brush of claim 49 wherein said hydrophobic polymer chain segments comprise repeat units derived from a hydrophobic monomer having a log P value of at least about 1.
- 54. The polymer brush of claim 49 wherein, in addition to said hydrophobic polymer chain segments, spacer molecules are also attached to the substrate surface, the ratio of said hydrophobic polymer chain segments to the sum of hydrophobic polymer chains segments and spacer molecules ranging from about 0.2 to about 0.8.
- 55. The polymer brush of claim 49 wherein at least a portion of the hydrophobic polymer chain segments are crosslinked to other hydrophobic polymer chain segments in the hydrophobic layer.
- 56. The polymer brush of claim 49 wherein the hydrophilic layer comprises water-soluble or water-dispersible polymer chain segments having groups for the attachment of a probe, wherein an end of each segment is attached to the hydrophobic layer, and further wherein said water-soluble or water-dispersible segments have a weight average molecular weight of at least 1,000.

- 57. The polymer brush of claim 56 wherein the water-soluble or water-dispersible segments have a weight average molecular weight of at least 1,000 but no more than 5,000,000.
- 58. The polymer brush of claim 49 wherein the hydrophilic layer comprises water-soluble or water-dispersible polymer chain segments having groups for the attachment of a probe, said segments comprising repeat units derived from a water-soluble or water-dispersible monomer having a log P value of less than about 1.
- 59. The polymer brush of claim 49 wherein the hydrophilic layer comprises water-soluble or water-dispersible polymer chain segments having groups for the attachment of a probe, said segments comprising repeat units derived from a water-soluble or water-dispersible monomer having a log P value of less than about 0.1.
- 60. The polymer brush of claim 58 wherein in an absolute difference between a log P value of the hydrophobic monomers and the water-soluble or water-dispersible monomers is at least about 1.
- 61. The polymer brush of claim 58 wherein in an absolute difference between a log P value of the hydrophobic monomers and the water-soluble or water-dispersible monomers is at least about 2.
- 62. The polymer brush of claim 49 wherein the hydrophobic layer comprises hydrophobic polymer chain segments, one end of each of said hydrophobic segments being attached to the substrate surface, and further wherein the hydrophilic layer comprises water-soluble or water-dispersible polymer chain segments having groups for the attachment of a probe, one end of each of said water-soluble or water-dispersible segments being attached to the hydrophobic layer, the ratio of water-soluble or water-dispersible segments to hydrophobic segments being less than about 1:1.

- 63. The polymer brush of claim 49 wherein the hydrophobic layer comprises hydrophobic polymer chain segments, one end of each of said hydrophobic segments being attached to the substrate surface, and further wherein the hydrophilic layer comprises water-soluble or water-dispersible polymer chain segments having groups for the attachment of a probe, one end of each of said water-soluble or water-dispersible segments being attached to the hydrophobic layer, the ratio of water-soluble or water-dispersible segments to a number of sites of attachment on the substrate surface being less than about 1:1.
- 64. The polymer brush of claim 49 wherein the hydrophilic layer has a dry thickness ranging from about 10 angstroms to about 2000 angstroms.
- 65. The polymer brush of claim 49 additionally comprising a probe attached to the functional groups for binding the molecule.
- 66. The polymer brush of claim 65 wherein said groups for the attachment of a probe are capable of attaching said probe without first being subjected to a chemical treatment to activate said groups for probe attachment.
- 67. The polymer brush of claim 65 wherein the molecule is a biological molecule and the probe is selected from the group consisting of nucleic acids, polypeptides, peptide nucleic acids, markers, cells, elastin, collagen, carbohydrates, enzymes, lipids, phospholipids, hormones, drug targets, phosphates, and metal ions.
- 68. The polymer brush of claim 65 wherein said functionalized groups are selected from the group consisting of hydroxy groups, amino groups, carboxylic acids, carboxylic acid derivatives, and thiols.

- 69. The polymer brush of claim 49 wherein the intermediate segment is substantially free of crosslinks to the intermediate segment of other polymer chains.
- 70. A sensor for binding a molecule in an aqueous sample in an assay, the sensor comprising:

a substrate surface;

a layer on the substrate surface comprising polymer chains having two termini and a water-soluble or water-dispersible intermediate segment between the termini, one terminus being free and the other terminus being bound to the substrate surface, the intermediate segment containing functionalized groups for the attachment of a probe for binding the molecule, the density of the functionalized groups being at least about 20 picomoles per square centimeter of substrate surface area;

spacer molecules bound to said surface, the ratio of polymer chains to the sum of polymer chains and spacer molecules ranging from about 0.02:1 to about 0.1:1; and,

a probe attached to the functional groups for binding the molecule, said probe having an average molecular diameter of at least about 50 angstroms.

- 71. The sensor of claim 70 wherein the probe has an average molecular diameter ranging from about 50 to about 250 angstroms.
- 72. The sensor of claim 70 wherein the probe has an average molecular diameter ranging from about 75 to about 150 angstroms.
- 73. The sensor of claim 70 wherein the ratio of polymer chains to the sum of polymer chains and spacer molecules ranges from about 0.04:1 to about 0.08:1.
- 74. The sensor of claim 70 wherein the polymer chains additionally comprise a hydrophobic segment between the water-soluble or water-dispersible intermediate segment and the substrate.

- 75. The sensor of claim 70 wherein the molecule is a biological molecule and the probe is selected from the group consisting of nucleic acids, polypeptides, peptide nucleic acids, markers, cells, elastin, collagen, carbohydrates, enzymes, lipids, phospholipids, hormones, drug targets, phosphates, and metal ions.
- 76. The sensor of claim 70 wherein the intermediate segment is substantially free of crosslinks to the intermediate segment of other polymer chains.
- 77. A method of preparing a polymer brush for binding a molecule in an aqueous sample in an assay, the method comprising:

forming a hydrophobic layer on a substrate surface having a dry thickness of at least about 50 angstroms; and,

forming a hydrophilic layer on said hydrophobic layer.

- 78. The method of claim 77 wherein said hydrophobic layer is formed by free radical polymerization.
- 79. The method of claim 78 wherein said hydrophobic layer is formed by first bonding a molecule, capable of initiating free radical polymerization, to a substrate surface at one or more points to form a derivatized surface.
- 80. The method of claim 79 wherein said derivatized surface is contacted with a composition comprising a hydrophobic monomer under free radical polymerization conditions to form said hydrophobic layer.
- 81. The method of claim 80 wherein said hydrophobic polymer layer is treated to render a fraction of the living free radical polymer chain ends incapable of re-initiating polymerization under free radical polymerization conditions.

82. The method of claim 81 wherein prior to said treatment, unbound hydrophobic polymer is removed.

141

- 83. The method of claim 82 wherein the derivatized surface is formed by bonding a nitroxide-containing molecule to the substrate surface, said treatment comprising heating the hydrophobic layer.
- 84. The method of claim 83 wherein the derivatized surface is formed by bonding an iniferter-containing molecule to the substrate surface, said treatment comprising exposing the hydrophobic layer to UV irradiation.
- 85. The method of claim 81 wherein said treated surface is contacted with a composition comprising a water-soluble or water-dispersible monomer under free radical reaction conditions to form said hydrophilic layer, said layer comprising polymer chains bound at one end to a hydrophobic polymer, said bound polymer chains comprising: (a) water-soluble or water-dispersible segments having a weight average molecular weight of at least about 1000; and (b) one or more functional groups capable of reacting with a probe selective for the molecule.
- 86. The method of claim 85 wherein the hydrophobic layer has a dry thickness of at least about 100 angstroms, 250 angstroms, 500 angstroms, 750 angstroms, 1000 angstroms, 1250 angstroms, 1500 angstroms or 2000 angstroms.
- 87. The method of claim 85 wherein the hydrophilic layer has a dry thickness ranging from about 10 angstroms to about 2000 angstroms, about 15 angstroms to about 1000 angstroms, or from about 25 angstroms to about 100 angstroms.
- 88. The method of claim 85 wherein the molecule is a biological molecule and the probe is selected from the group consisting of nucleic acids, polypeptides, peptide nucleic acids, markers, cells, elastin, collagen,

carbohydrates, enzymes, lipids, phospholipids, hormones, drug targets, phosphates, and metal ions.

- 89. The method of claim 85 wherein the water-soluble or water-dispersible segments of the polymer chain are substantially free of crosslinks to the water-soluble or water-dispersible segments of other polymer chains.
- 90. A method of preparing a polymer brush for binding a molecule in an aqueous sample in an assay, the method comprising:

bonding a molecule, capable of initiating free radical polymerization having living-type kinetics to a surface of a substrate at one or more points to form a derivatized surface;

contacting said derivatized surface with a composition comprising a hydrophobic monomer under living free radical reaction conditions to form a bound, hydrophobic polymer layer;

treating said hydrophobic polymer layer to render a portion of living free radical polymer chain ends incapable of re-initiating polymerization under free radical polymerization conditions; and,

contact said treated layer with a composition comprising a water-soluble or water-dispersible monomer under free radical reaction conditions to form polymer chains bound at one end to a hydrophobic polymer, said bound polymer chains comprising: (a) a water-soluble or water-dispersible segments having a weight average molecular weight of at least about 1,000; and (b) one or more functional groups on said bound polymer chains that are capable of reacting with a probe selective for the biological molecule.

- 91. The method of claim 90 wherein prior to said treatment, unbound hydrophobic polymer is removed.
- 92. The method of claim 91 wherein the derivatized surface is formed by bonding a nitroxide-containing molecule to the substrate surface, said treatment comprising heating the hydrophobic layer.

- 93. The method of claim 92 wherein the derivatized surface is formed by bonding an iniferter-containing molecule to the substrate surface, said treatment comprising exposing the hydrophobic layer to UV irradiation.
- 94. The method of claim 93 wherein the surface-bound iniferter initiator has the formula:

wherein: C is a moiety on the surface of the substrate; L is a linker group capable of bonding to at least one C moiety; z is 0 or 1; q, r and t are independently 0 or 1, provided the sum of q + r + t is at least 1; Y is a residue capable of initiating free radical polymerization upon homolytic cleavage of the Y-S bond; S is sulfur; and, G is a nitrogen or an oxygen heteroatom.

95. The method of claim 94 wherein the surface-bound iniferter initiator has the formula:

wherein N is nitrogen, and R¹ and R² are independently selected from hydrogen, hydrocarbyl and substituted hydrocarbyl.

96. The method of claim 90 wherein the hydrophobic layer has a dry thickness of at least about 100 angstroms.

- 96. The method of claim 90 wherein the hydrophobic layer has a dry thickness of at least about 100 angstroms.
- 97. The method of claim 90 wherein the hydrophobic layer has a dry thickness of at least about 500 angstroms.
- 98. The method of claim 90 wherein the water-soluble or water-dispersible polymer chain segments have a weight average molecular weight of at least 1,000 but no more than 5,000,000.
- 99. The method of claim 90 wherein said functional groups are selected from the group consisting of hydroxy groups, amino groups, carboxylic acids, carboxylic acid derivatives, and thiols.
- 100. The method of claim 90 wherein the molecule is a biological molecule and the probe is selected from the group consisting of nucleic acids, polypeptides, peptide nucleic acids, markers, cells, elastin, collagen, carbohydrates, enzymes, lipids, phospholipids, hormones, drug targets, phosphates, and metal ions.
- 101. The method of claim 90 wherein the intermediate segment is substantially free of crosslinks to the intermediate segment of other polymer chains.
- 102. A polymer brush for binding a molecule in an aqueous sample in an assay, the brush comprises a substrate surface having a polymer layer thereon, said polymer layer comprising a first hydrophobic layer attached to the substrate surface, and a second hydrophilic layer attached to the hydrophobic layer containing groups for the attachment of a probe for binding the molecule, said brush being characterized in that, upon being immersed in a 10 mmolar sodium hydroxide solution for about 15 minutes, the polymer layer thickness is reduced by less than about 40%.

- 103. The polymer brush of claim 102 wherein the polymer layer thickness is reduced by less than about 20%.
- 104. The polymer brush of claim 102 wherein the polymer layer thickness is reduced by less than about 10%.
- 105. The polymer brush of claim 102 wherein the hydrophobic layer has a dry thickness of at least about 100 angstroms.
- 106. The polymer brush of claim 102 wherein the hydrophobic layer has a dry thickness of at least about 1000 angstroms.
- 107. The polymer brush of claim 102 wherein the hydrophilic layer has a dry thickness ranging from about 10 angstroms to about 2000 angstroms.
- 108. The polymer brush of claim 102 wherein the hydrophilic layer comprises water-soluble or water-dispersible intermediate segments having a weight average molecular weight of at least about 1000, and one or more functional groups capable of reacting with a probe selective for the molecule.
- 109. The polymer brush of claim 108 wherein said functionalized groups are selected from the group consisting of hydroxy groups, amino groups, carboxylic acids, carboxylic acid derivatives, and thiols.
- 110. The polymer brush of claim 109 wherein the molecule is a biological molecule and the probe is selected from the group consisting of nucleic acids, polypeptides, peptide nucleic acids, markers, cells, elastin, collagen, carbohydrates, enzymes, lipids, phospholipids, hormones, drug targets, phosphates, and metal ions.